"THE COMPOSITION, YIELD AND NUTRITIONAL VALUE OF WHEAT FROM THE BRETON PLOTS"

J. A. Carson, B.Sc.

University of Alberta

April, 1958

THE UNIVERSITY OF ALBERTA

"THE COMPOSITION, YIELD AND NUTRITIONAL VALUE OF WHEAT FROM THE BRETON PLOTS"

A DISSERTATION

SUBMITTED TO THE FACULTY OF GRADUATE STUDIES

IN PARTIAL FULFILMENT OF THE REQUIREMENTS FOR THE DEGREE

OF MASTER OF SCIENCE

FACULTY OF AGRICULTURE

DEPARTMENT OF SOIL SCIENCE

by

J. A. CARSON, B.Sc.

EDMONTON, ALBERTA

APRIL, 1958

ABSTRACT

The object of this investigation was to determine the effect of crop rotations and fertilization on the yield, chemical composition and nutritive value of grain grown on the Breton Plots. The nutritive value of wheat was determined by using rats as laboratory animals for the feeding trials.

The sulfur content of wheat was increased slightly by the application of fertilizers supplying sulfur while the remaining mineral constituents were not increased or decreased consistently by fertilizer treatments.

There was a marked increase in the protein content of wheat following legumes in rotation. The application of fertilizers to wheat resulted in a slight increase in the per cent protein although results were variable. The increase in protein content of wheat following a legume crop resulted in a significant increase in rat gain compared to rats fed wheat after fallow. Manure application to wheat after fallow also caused a significant increase in nutritive value as measured by rat gains. Wheat following fertilized legumes gave the highest nutritive value of any wheat grown on sulfur deficient soil of the Breton Plots.

ACKNOWLEDGEMENTS

The writer wishes to express his appreciation to Dr. C. F. Bentley, Dr. J. D. Newton, Dr. J. A. Toogood and Mr. J. A. Robertson for assistance throughout the course of this investigation and for criticism of the manuscript.

The author is also indebted to other members of the Department of Soil Science who have cooperated and thus made this research investigation a pleasant task.

Acknowledgement is made to Dr. J. P. Bowland and the other members of the Department of Animal Science for making available facilities necessary for conducting the rat feeding trials. Thanks are also extended for helpful suggestions in planning the feeding trial investigation.

Appreciation is also expressed to the Department of Soil Science for financial assistance during the course of this investigation.

TABLE OF CONTENTS

LIST OF TABLES AND PLATES

THE COMPOSITION, YIELD AND NUTRITIONAL VALUE OF WHEAT FROM THE BRETON PLOTS

INTRODUCTION

The Grey Wooded Soil Zone covers about 100 million acres, or two-thirds of Alberta. However, only a small portion of this is considered to be potential agricultural land. The potentially arable Grey Wooded area has been estimated to comprise about 10 - 25 million acres north and west of Edmonton. It is evident that until exploratory surveys now in progress are completed, we shall not know the agricultural potential of this large portion of Alberta.

The arable soils of the Grey Wooded Zone of northern Alberta and the irrigable land of the south appear as possibilities for future agricultural development. Some agriculturists are of the opinion that at present settlement in the Grey Wooded Zone should go no further, and that more specialized farming should be encouraged on presently developing irrigation schemes. Others argue that the cost of development of irrigated land is prohibitive and that settlement on Grey Wooded soil should be encouraged. However, within the next few decades both areas are likely to be important. For this reason, and to assist those already farming Grey Wooded soil, it is desirable to determine the best use and management of these soils.

Cropping practices that include rotations and fertilizer treatments can bring about changes in crop yield and composition and at the same time they can physically affect Grey Wooded soils. Evidence to support this claim is contained in the bulletin, "Wooded Soils and Their

Management", by Newton et al. (43). Studies at the Breton Plots, reported in that bulletin, have shown that rotations which include a legume and suitable fertilization bring about yield increases and also increase the protein content of grains and hays produced. Thus, crop production on sulfur deficient Grey Wooded soils has been substantially improved when suitable fertilizers are included in a rotation containing legumes.

Since protein is an important feed constituent, and since soil management has affected this nutrient in Breton crops, it was desired to know what, if any, changes there have been in the nutritive value of the crops produced there. In order to evaluate the effect of fertilizer treatments on forage crops, feeding trials using rabbits as test animals have been conducted at the University of Alberta since 1951 (13). Those experiments have shown that the nutritive value of legume hays from plots receiving recommended fertilization is higher than that of hay from adjacent unfertilized areas. The tests have proven to be very satisfactory for analyses of the feed value of forages and are thought to reveal differences which are important to farmers on similar soils.

The present investigation was undertaken in an endeavor to determine the effects of crop rotations and fertilization on the yield, chemical composition and nutritive value of grain grown on the Breton Plots.

REVIEW OF LITERATURE

A. Effects of Cropping Practices on Composition of Grain Crops.

1. Effect of Rotations - Cropping practice introduces two separate aspects of management that may change the yield and chemical composition of the succeeding crops. One aspect is the crop sequence employed and the second is the application of fertilizers. Since these practices are important, not only to the farming population, but also to the research investigator, some recent publications related to these matters will be reviewed.

The use of long term experimental plots is a good method of studying the effect of crop sequences, or rotation, on the yield of grain crops. The world's oldest experimental plots are at Rothamsted Experimental Station. Other similar long term experimental plots have been established in the United States and Canada. Miller (35) has reviewed some of the existing long term experiments in the United States. He found that in general grain crops grown after legumes gave higher yields than grain grown continuously. Experimental plots that have been established and continued for several years are a very satisfactory means of investigating the effects of crop rotation on the yield of grain.

The effects of alfalfa, grass and wheat crop residues on yield of grain which followed were studied in Manitoba. Ellis (11) reports that grass-sod lowered the yield of grain, both as first and second crop after breaking, compared to a wheat-wheat sequence. However, the decrease

in yield was counteracted when an alfalfa-grass mixture was used in place of a pure grass stand. Similar results were reported by Hedlin et al. (18), also in Manitoba. They report yields of the first wheat crop after alfalfa, grass and wheat residues as follows: 34.4 bushels per acre following alfalfa; 25.6 bushels per acre after brome grass; and 31.4 bushels per acre after wheat residue. This depression in yield after brome grass and wheat residue was more pronounced in the second wheat crop after ploughing down the residue. They also stated that ploughing down sweet clover or burning the wheat straw increased the yield of the second wheat crop following these treatments compared to the control crop of wheat on stubble. Alfalfa or an alfalfa-grass mixture is the forage that is likely to have the greatest effect on maintenance of yield in subsequent wheat crops.

Hill (19) reports the yield effect of rotations at three experimental station operated in Western Canada over the past half century. He states that results from the Brandon, Manitoba station indicate that wheat yields have been maintained in a fallow-wheat rotation, while an adjacent 6 - year rotation containing three years of hay has had higher yields. Yield results from the Indian Head station quoted by Hill, show that wheat yields after fallow have been maintained at the original level, while yield of wheat after wheat has steadily declined with the passing years. Both rotations were adversely affected by weeds. Hill's data from Lethbridge station show wheat yields after fallow have been maintained at a higher level than: 1) those on continuous wheat plots; 2) 9 - year rotation which includes corn and hay; 3) 10 - year rotation which includes corn and alfalfa for seed. The longer rotations were stated to be less profitable because of the comparatively low monetary value of hay

and alfalfa. However those conclusions may not be warranted because hay prices have increased considerably in recent years. This work from these Western Canadian experimental stations shows that yields of wheat following summerfallow can be maintained at a higher level than when wheat follows other grains or hay in rotations.

Bains (5) reports an investigation in which phosphorus uptake by grain and the subsequent utilization of the grain phosphorus by rats were studied. Four fertilizer treatments were used on the grain: check; green manure; green manure plus phosphate fertilizer; and phosphate fertilizer alone. The respective effects of these treatments on the phosphorus content of grain produced were: increased phosphorus content of grain grown after green manure; an additional increase in phosphorus content of grain from green manure plus phosphate fertilizer. The feeding trial showed that the same amount of phosphorus was retained by the rats when fed grain from plots treated with green manure plus phosphate fertilizers as was retained when they were fed grain from plots treated with phosphate fertilizers. Phosphorus retention by the rats was lowest when they were fed grain from untreated plots. The explanation offered for these results was that the form of phosphorus in the grain depends on the chemical form of phosphorus in the soil. The phytin phosphorus content of grain treated with phosphorus fertilizer was the same as that treated with phosphate fertilizer plus green manure, and this may be the form of grain phosphorus utilized by rats. This study has shown that soil management and fertilization can affect the phosphorus content of a grain and the form in which phosphorus is present in grain.

Work reported from Western Canada suggests that legumes without fertilization influence the protein content of the wheat crop in rotations.

Ellis (11) and Hedlin et al. (18) reporting results from Manitoba found protein content of wheat grown after breaking grass sod lower than that of grain grown after fallow. However wheat grown after legumes had a higher protein content than the wheat on fallowed land. Newton et al. (43) reporting results from Breton found that wheat after legumes averaged 12.6 per cent protein compared to 10.1 per cent in wheat after fallow. Since protein is an important constituent of grain, such an increase, if it can be brought about by a soil management program, may represent an important benefit.

Cropping practices that include legumes may increase yield and affect the chemical composition of succeeding grain crops; these practices may also improve the soil for future production.

2. Effects of Fertilizers - Profitable direct returns from fertilizer use are very common and will not be reviewed here. However, an indirect return from fertilizer may be an improvement in the protein or mineral composition of the crop produced.

The application of nitrogen fertilizer has caused increases in the nitrogen content of some crops. Russell et al. (56), working with brome grass in Nebraska, found a linear relationship between the rate of nitrogen fertilization and the nitrogen content of the grass. They also found that the per cent nitrogen at all rates of application was higher during early stages of crop growth than later in the growing season. For this reason it is desirable to apply high rates of nitrogen fertilizer and to cut hay before flowering.

Fertilizers that include phosphorus and/or potassium with nitrogen may not cause an increase in protein content of grains and alfalfa. Bains (6)

found that phosphorus applied as superphosphate produced a low protein grain, but the application of ammonium sulfate plus superphosphate produced a high protein wheat. Similarly, Murphy (40) working in Oklahoma found that the protein content increased as the nitrogen in the fertilizer increased, while the application of phosphorus alone, with nitrogen, or with potassium, decreased the protein content. The foregoing results are not in complete agreement with those obtained by Huppert (21) in Germany. He found that light applications of nitrogen fertilizer decreased the protein content of wheat, while heavy applications increased it. Only relatively heavy applications of nitrogen fertilizer increased the protein content of feedstuffs.

There are other factors affecting the per cent nitrogen of wheat besides the direct influence of fertilization. The time of application of nitrogen fertilizers was studied by Peterson (48) in Utah. He found that spring applications were more effective than fall applications in increasing the protein content of winter wheat. A similar study by Long et al. (28) showed that a late application of nitrogen fertilizer to wheat grown in Tennessee increased the per cent protein more than the earlier, March or November, applications. Another factor that Ellis (11) studied in Manitoba was the effect of seasonal variations on protein content of wheat. He found that even though the seasonal variation was great, there was a slight increase in protein content due to the application of nitrogen fertilizers. He also found a slight tendency for phosphate fertilizers to decrease the protein content. Since the time of application of fertilizers and seasonal variations cause changes in protein content of grains, it is difficult to evaluate the results of experimental data for one year.

The effects of fertilization on the phosphorus content of grain and legume crops has been studied in Eastern Canada and Wisconsin. MacLean et al. (30), working in Quebec, determined the phosphorus content of grain grown on ten different soil types and fertilized with phosphate fertilizers. They found from greenhouse experiments that the phosphorus content of oats was increased slightly as a result of fertilization on seven of the ten soil types. Results of phosphorus application to an alfalfa crop on these same soils showed very inconsistent results. Wedin et al. (61) in Wisconsin obtained results differing from those obtained in Eastern Canada. They found that phosphorus fertilization increased the phosphorus content of alfalfa and Ladino clover. Widely separated locations may give different results even though the same fertilizer and the same crop are used.

The utilization of phosphorus may be affected to some extent by the fact that phosphorus absorbed by plants is converted to organic phosphorus compounds (10). Since this conversion may be slow, the uptake may be restricted and the period of absorption extended so that phosphorus becomes "fixed" before it can be utilized (38). Mitchell (37), using "radioactive tracer techniques" , has found that wheat uses more fertilizer phosphorus in the early stages of crop growth than later in the season. He found in addition that the uptake of fertilizer phosphorus increased with increasing increments of fertilizer up to 24 pounds of phosphorus per acre. The application of fertilizer phosphorus may increase the phosphorus uptake by grain crops.

Factors other than phosphate fertilization have been found to change the phosphorus content of crops. Russell et al. (56), for example,

working in Nebraska found that nitrogen fertilization increased the phosphorus content of bromegrass early in the growing season but at harvest the p r cent phosphorus was the same as that of the check plot. Rennie (53), in Saskatchewan, found that nitrogen fertilization increased the phosphorus content of grain when cut as wheat forage but when left to maturity the per cent phosphorus decreased becoming lower than the unfertilized grain. Soil type and climate as factors causing variation in per cent phosphorus of grain were also investigated by Rennie. He found greater changes in the phosphorus content of wheat between soil types and between seasons than changes due to fertilization. It would appear from these reports that, to arrive at any general conclusion concerning the effects of fertilization on phosphorus content of crops, it would be necessary to have several years' results at a single location.

Many more factors have an effect on the phosphorus content of grain. Two of these factors are the presence in the soil of large amounts of calcium (26) and silicate (3). In conclusion it may be stated that in Vandecaveye's (60) review only 50 per cent of the reports he considered showed fertilization to have increased the phosphorus content of grains.

The effect of fertilization on potassium, sodium, calcium, and magnesium has been investigated extensively in Central United States (62, 12, 34). The results reported by these workers are variable, but Vandecaveye (60) concluded that in general fertilization caused inconsistent changes in these mineral constituents and that these changes were more dependent on soil types and season.

The effect of sulfur fertilization on the sulfur content and yield of grains and legumes has been studied extensively in regions of low soil sulfur. Powers (49) states that a well drained soil in regions where the precipitation is relatively free of industrial residues may occasionally

be low in sulfur. Sulfur fertilization of these sulfur deficient soils may increase the yield and sulfur content of some crops. Rendig (51), working in California on a sulfur deficient soil, found that the application of 200 pounds per acre of gypsum increased the sulfur content of alfalfa from 0.16 per cent to 0.33 per cent. Peters (46) investigated the sulfur content of grain grown on sulfur deficient soil on the Breton plots. He found that fertilizer treatments which included sulfur caused large yield increases and the sulfur content of these crops was considerably higher than that of unfertilized crops. Hoff (20) and Gareau (13) have also reported that sulfur fertilization increases the sulfur content of Alberta crops on Grey Wooded soils. Hoff also found some increase in sulfur content of fertilized crops grown on soils not sulfur deficient. Relatively large increases in sulfur content of grains as a result of sulfur fertilization are limited to soils low in sulfur.

A study of the sulfur containing amino acids of wheat protein made by Kasting (24) found that wheat grown on the Breton plots had a cystine content proportional to the total sulfur content. This increase in cystine was not correlated with loaf volume in baking tests when wheat followed clover, but a positive correlation did result with continuous wheat. Sulfur fertilization on the Breton plots has not only increased yield (43) but has improved the quality of the grain crops.

B. Feeding Trials with Laboratory Animals

Many analytical procedures have been tried to measure the nutritive value of grains and hays, but the most reliable method is animal feeding trials. Since the ultimate use of grains and hays is in the living animal, in vitro tests of quality may be artificial while in

vivo studies are more realistic.

Some experiments have been conducted using cattle to evaluate the nutritive value of hays. Lynd et al. (29) in Oklahoma used paired steers in a study of the effect of fertilizer treatments on the nutritive value of a grass-legume mixture. Their technique has possibilities in evaluating the nutritive values of pasture production. Crampton et al. (9) used steers to determine the effects of fertilization on the carrying capacity of pasture and on the rate of animal gain. They used a complete fertilizer, and also considered the economics of fertilizer use. The data, a four year average, showed a doubling of both carrying capacity and beef gain per acre due to fertilization. The increase in beef gain was more than sufficient to pay the cost of the fertilizer. Using cattle in experiments requires several years to obtain sufficient data to draw sound conclusions, and the amount of feeding material required is large making such experiments costly.

The sheep is a small ruminant with low feed requirements and is therefore convenient to use in experiments intended to determine the comparative feeding value of forages. In using lambs, Weir (63) and Smith et al. (58) found hay from phosphate fertilized plots had a higher nutritive value than unfertilized hays. These experiments with sheep required considerable replication with the result that considerable quantities of experimental feeds were required and overall costs of the experiments were rather large.

Rabbits have a high ability to digest forage even though they are not ruminants (13). Since the digestive processes of rabbits are quite similar to those of ruminants (13) and because their feed requirements

are comparatively low they are convenient to use for evaluating the nutritive value of forages. Koehler et al. (25) using rabbits found that the application of magnesium carbonate to an alfalfa crop in Missouri improved the feeding value of the forage. Gareau (13) used rabbits to determine the effect of fertilization on the nutritive value of Alberta hays. He found that on sulfur deficient soils, including the Breton plots, sulfur fertilization increased the protein content of the feeds and this was accompanied by higher nutritive value. Although not a ruminant, Gareau concluded,"the rabbit is a very valuable animal for feeding trials of this nature".

Other small laboratory animals such as the chick (47) and the guinea pig (61) have been used to evaluate the nutritional value of feeds.

There has been a relatively large number of investigations conducted using the rat as a laboratory animal to determine the nutritive value of grains. This animal is especially useful when protein and its constituents are a major consideration, since the amino acid requirements of rats are very similar to those of swine (31).

The limiting amino acids of rice and peas have been determined using the rat as a test animal. Harper et al. (17) found that, for the rat, polished rice was deficient in lysine and threonine. After supplementation of rice with lysine, no further growth increase was obtained until a mixture of all the essential amino acids was added. The first limiting amino acid of field peas is methionine as Murray (41), Murray et al. (42)and Woods et al. (64) found with rat feeding trials. In addition, Woods et al., working in Idaho with peas, found that supplementation with 0.3 per cent methionine gave the best rat growth.

Rat investigations have been conducted to determine the limiting amino acids of wheat and wheat flour for rats. These animals grew better when they were fed white flour supplemented with lysine and valine (27). Rosenberg et al. (55) working with wheat flour in Delaware reported that lysine was the only limiting amino acid. Mitchell et al. (36) established that, for wheat, lysine was the first limiting amino acid. However, when lysine was supplemented in sufficient quantitites the addition of trytophan gave a further growth increase. Sure (59) also found that whole wheat was limiting in lysine but in addition it was limiting in valine and threonine. These investigations have established that for the rat the main limiting amino acid of flour and wheat is lysine and that other amino acids are limiting when lysine is supplemented.

The limiting amino acid in a particular grain is important when investigations are conducted to ascertain at what level they should be supplemented. Rosenberg et al. (54) determined the level of lysine necessary to supplement flour in order to attain good rat growth. They concluded that flour supplemented with 0.8 per cent lysine would produce growth equal to that obtained with a stock diet which was well balanced, containing 21.5 per cent protein and 1.1 per cent lysine. Hutchinson et al. (22) found that supplementing white bread with 0.25 per cent lysine gave optimum growth in rats. They also reported that increasing the content of a single amino acid may lead to a reduced rate of growth by reducing the utilization of the other amino acids. Harper et al. (16) reported on the growth retarding effect of amino acid unbalance. They found that excess dietary leucine retards rat growth on diets deficient in isoleucine. They concluded that leucine in the diet increases the requirement of isoleucine. The foregoing results are in general agreement

with data by Morrison (39) and Maynard et al. (31) in that, for the rat, lysine and methionine are the amino acids most likely to be deficient in hard red spring wheats.

Renner et al. (52) studied the effect of various fertilizers on the amino acid content of wheat grown on the Breton plots of Alberta. Only fertilizers containing sulfur increased the nine essential amino acids of wheat grown in rotation with legumes. There was no evidence of fertilizer treatments affecting the proportions of amino acids in wheat from the wheat-fallow system. A comparison of the grain from the check plots of the two cropping systems shows that, even though there was an increase in the quantity of protein in wheat after legumes, there was a decrease in quality as measured by the proportion of these nine essential amino acids. Moreover the proportions of the nine essential amino acids in fertilized wheat grown after legumes was the same, not higher, than the proportions in the wheat grown after fallow. McElroy et al. (33) in Alberta found marked differences in rat growth attributable to differences in total protein content of wheat grown there. Rats fed wheat containing 17.3 per cent protein gained 70 per cent more than rats fed wheat containing 8.9 per cent protein. It was later found that these results were applicable to pigs (32). The results by Renner et al. (52) suggest the possibility that crop rotation and fertilization at Breton are causing changes in the proportions of amino acids in wheat which may affect rat growth. A change in the protein content of the diet may affect both the digestibility of the diet and gains made by rats, since the rate of protein intake by rats closely parallels the body requirements (14).

There has been a considerable amount of investigation carried out which suggests that rats would be suitable animals to use in experiments intended to determine whether crop rotations and fertilizer applications are causing differences in nutritional value of grain from the Breton plots.

MATERIALS AND METHODS

A. Source of Materials

Breton Experimental Plots - Plots 1 to 11 at Breton were established in 1930 on a Grey Wooded soil, about two miles south-east of the town of Breton. Breton is approximately 70 miles south-west of Edmonton. Treatments used and the results obtained, accompanied by a full description of these plots, are outlined by Newton et al. (43). Several technical reports relating to the Breton plots as well as ones on similar sulfur deficient soils of Alberta are available (20, 13, 46).

This investigation was primarily concerned with six fertilizer treatments on each of, wheat after fallow, and wheat after legumes. In 1956 the areas involved were, series E (wheat after fallow) and series B (wheat after legumes), while in 1957 the areas were again series E and series C (wheat after legumes). Under the system of fertilization employed on the Breton plots, fertilizers were applied in 1956, thus there was only a residual effect on the 1957 crop. The experimental material used in this investigation was obtained from the plots listed in Table I.

The wheat for the feeding trials was cut at normal harvest time after sampling for yield determinations. It was desirable to obtain approximately 30 pounds of wheat from each of the six plots. The wheat was cut with a binder, cured and threshed. After removal of all weeds and other grains, the wheat was ground in a Wiley mill to pass a 20 mesh sieve. Representative samples for chemical analyses were placed in labelled four ounce bottles. The remainder was retained for rat feeding trials.

The barley from Breton was included in the chemical analyses but was not used in the rat feeding trials. The barley in 1956 was grown the first year after legumes (Series B - west half). The two 1957 barley

Table No. I

FERTILIZER TREATMENTS AND PLOTS USED FOR EXPERIMENTAL MATERIAL

Plot Number	Fertilizer Material	Symbol used Hereafter	Rates/acre	Nutrients supplied - lbs/acre/year N	P_2O_5	K_2O	S
1 & 5	None	Check	-	-	-	-	-
2	Manure	M	20 tons - every 5 yrs.	40[2]	20[2]	40[2]	4[2]
3	16-20-0 + KCl	NPKS	100 lbs. - every 2 yrs. 50 lbs. - every 2 yrs.	16	20	30	14
4	21-0-0	NS	56 lbs. - every 2 yrs.	16	-	-	14
6	Lime	L	4850 lbs. - every 10 yrs.	-	-	-	-
9	16-20-0 + manure	NPS + M	100 lbs. - every 2 yrs. 20 tons - every 5 yrs.	56	40	40	18

1. These are the rates and materials currently applied. The quantities of fertilizers used varied but the kind of nutrients applied has been the same over the years.

2. Plant nutrients in manure vary with the kind and age of manure.
The manure is applied to the second year legume crop, the year before the wheat after legume crop.
The manure in the wheat fallow system was applied in 1952 and 1957.

crops were grown the first and third year after legumes. The plots used were the same as those used for the wheat crops listed in Table 1.

Information on analytical work done by the author during 1955-1956 is included in the appendix of this report. This material is not directly related to the present investigation but is included in the appendix as a formal and permanent record of that work.

B. Analytical Procedures

Total nitrogen determinations were by the Kjeldahl-Gunning method (44) using mercury as catalyst. Sodium thiosulfate was added in the 40 per cent sodium hydroxide to precipitate the mercury. The ammonia distillate was collected in 4 per cent boric acid and titrated with N/14 sulfuric acid (44,50). The nitrogen content of the urine was determined on a 20 ml. aliquot from the samples collected. The procedure for their collection is described later under C. Rat Feeding Trials.

The grains from Breton as well as grains from the 1956 cooperative trials were analyzed for sulfur, potassium, calcium, magnesium, sodium and phosphorus. The grain samples were ashed by a modification of the procedure outlined by Gieseking et al. (15) and presented by Hoff (20). The details of the procedure used are given in the appendix. The extract from this digestion was used to determine the sulfur, sodium, potassium, calcium and magnesium content of the samples. Sulfur was determined turbidimetrically according to the method outlined by Hoff (20). Sodium and potassium were determined with a Model D.U. Spectrophotometer using a slight modification of Gareau's (13) procedure, the details of which are given in the appendix. Calcium and magnesium were determined using the method outlined in detail by Pawluk (45). For the phosphorus determination, the grain samples were ashed by the magnesium nitrate method given

in A.O.A.C. (44) and determined colorimetrically with a Bausch and Lomb Monochromatic colorimeter, using the ammonium meta-vanadate method outlined by Gareau (13).

The gross energy of the rat feed and feces was determined using a Parr Oxygen Bomb Calorimeter. The use of an oxygen bomb calorimeter is recommended by Bell (7) for the determination of energy in material of this nature. The details of the procedure are outlined in the Parr manual (4).

Yield data from the Breton plots weredetermined from a four square yard sample per treatment. The grain was weighed in grams and the yield per acre calculated. The cooperative trial yield data weredetermined from ten square yard samples per treatment.

C. Rat Feed Trials

1. Supplements in Feeding Trials - There are minimum amounts of vitamins and minerals necessary for good rat growth (8, 23). For this reason a mineral supplement recommended by Jelinek et al. (23) was added to the grains used in this experiment. A vitamin supplement as used by Sibbald et al. (57) was also used. Lysine was added at 0.9 per cent of the ration which is slightly higher than that suggested by Rosenberg et al. (54). Details concerning these materials and of the procedure for their addition are given in the appendix.

The diet containing grain, vitamins, minerals and lysine was mixed one week prior to commencement of the feeding trials.

2. Procedure of Rat Feeding Trials - Albino weanling rats of the Sprague-Dawley strain were allotted to individual cages, housed in the animal room of the Department of Animal Science. After the animals had been assigned to groups and cages, they were fed their assigned feeds during a preliminary acclimatization period of one week. The rats weighed approximately 30 to 40 grams when they were weaned. After the acclimati-

zation period, the animals were transferred to metabolism cages for one week, during which time feed consumption was recorded. The rats were fed in Joy food cups to prevent wastage. These cages were constructed with a removable tray, which was tapered toward the centre for collecting urine in 10 ml. of 50 per cent sulfuric acid. A screen was placed over the tray to collect the feces separately from the urine. The cages were sprayed with 2 per cent boric acid in 95 per cent alcohol prior to the experiment in order to prevant losses of the ammonical nitrogen. The cages were rinsed with hot distilled water at the conclusion of the metabolism period. The total urine collected and cage washings were made up to 500 ml. The nitrogen content of the urine was determined by using a 20 ml. aliquot for the analysis. The feces were collected every morning during the metabolism period and oven dried at 105 C. At the conclusion of the metabolism period the feces were weighed, ground to pass a 20 mesh sieve, and stored in four ounce jars for analysis.

The animals were weighed at the beginning of acclimatization period, at the beginning of metabolism period, and at the conclusion of the experiment.

RESULTS AND DISCUSSION

A. 1956 Breton Grains

1. Yield and Chemical Composition - The yield data and the results of chemical analyses for 1956 Breton wheat after legumes and wheat after fallow are reported in Table 2.

The yield of wheat after legumes shows a marked increase due to the application of sulfur containing fertilizers and manure. However, this trend is not evident in wheat after fallow. Similar results have been obtained in extended experiments on sulfur deficient Grey Wooded soils at Cheddarville (1) and Breton (2). The manure treatment in these extended tests has resulted in nearly equal yield increases on both wheat after fallow and wheat after legumes. This is not evident in the Breton data reported in Table 2 and may be because the manure had not been applied since 1952. From the 1956 data it appears that the preceding legume crop and fertilization has increased wheat yields the most.

The chemical composition of wheat has been affected slightly by cropping practice and fertilization. The nitrogen content showed the greatest increase as a result of these practices. Fertilization has increased the per cent nitrogen more in wheat after legumes than in wheat after fallow. Moreover, the nitrogen content of wheat after legumes on all plots is higher than that of wheat after fallow. This agrees with the findings of Newton et al. (43). They report that the cropping system ? influenced the protein content of wheat at Breton more than the direct application of fertilizers. The phosphorus content of wheat after legumes is slightly higher than that of wheat after fallow. The phosphorus content of wheat after legumes shows a slight decrease due to the application of nitrogen and sulfur containing fertilizers, while in wheat after fallow

Table No. 2

YIELD AND CHEMICAL COMPOSITION OF 1956 BRETON WHEAT

Treatment	Yield bu/ac.	%N	%P	%K	%S	%Ca	%Mg	%Na
Wheat after legumes								
Check	13.8	2.12	0.50	0.50	0.12	0.02	0.14	0.010
L	13.4	2.46	0.53	0.50	0.09	0.04	0.14	0.008
M	34.8	2.41	0.46	0.51	0.10	0.03	0.11	0.007
NS	38.7	2.37	0.44	0.45	0.16	0.04	0.11	0.009
NPKS	33.6	2.34	0.49	0.46	0.13	0.02	0.11	0.008
NPS + M	42.3	2.32	0.46	0.50	0.14	0.03	0.08	0.010
Wheat after fallow								
Check	8.4	1.97	0.41	0.51	0.13	0.03	0.14	0.016
L	10.8	2.11	0.49	0.52	0.12	0.03	0.14	0.014
M	8.3	2.10	0.49	0.51	0.12	0.01	0.09	0.013
NS	7.1	1.99	0.50	0.52	0.13	0.02	0.09	0.012
NPKS	10.3	1.88	0.46	0.52	0.12	0.03	0.10	0.008
NPS + M	14.5	2.12	0.48	0.53	0.12	0.04	0.13	0.009

Note: Yields based on a 4 square yard sample; chemical analyses averages of duplicates and expressed on oven dry basis.

there appears to be a slight increase in per cent phosphorus from all the fertilizer applications. These data agree with results reported by Rennie (53) who found that fertilization of wheat after legumes decreases the phosphorus content. Sulfur and nitrogen fertilization increased the sulfur content of wheat after legumes slightly, while on wheat after fallow the sulfur content has remained relatively unchanged. Peters (46) and Hoff (20) found that these fertilizers increased the sulfur content of both wheat after legumes and wheat after fallow. The 1956 results show that the application of sulfur plus nitrogen fertilizers decreased the sodium and magnesium content of both wheat crops. Of the mineral constituents determined in the wheat crop, nitrogen was affected most, both by fertilizer application and the preceding legume crop.

The yield and chemical composition of barley grown the first year after legumes are reported in Table 3. The yield of the barley crop was very poor which is similar to the results of wheat after fallow reported in Table 2. However, the barley yields show a greater increase due to the application of fertilizers than wheat after fallow and less than the yield increases of wheat after legumes. The mineral constituents of barley reported in Table 3 were increased slightly by the application of fertilizers, with the nitrogen and sulfur content showing the greatest increase. Manure application on this barley crop has not only given the largest yield but has increased nitrogen, potassium, sulfur and calcium content of the barley. Magnesium and sodium decreased with the manurial treatment. The application of fertilizers to barley has resulted in an increase in yield, a decrease in per cent phosphorus and sodium and no effect on per cent calcium and magnesium.

Table No. 3

YIELD AND CHEMICAL COMPOSITION OF 1956 BRETON BARLEY

Treatment	yield bu/ac.	%N	%P	%K	%S	%Ca	%Mg	%Na
First year barley after legumes								
Check	3.8	1.90	0.52	0.56	0.09	0.04	0.09	0.017
L	1.5	2.12	0.62	0.59	0.07	0.04	0.08	0.007
M	22.8	2.10	0.52	0.59	0.11	0.06	0.04	0.013
NS	20.8	2.06	0.39	0.54	0.15	0.04	0.11	0.013
NPKS	16.7	2.15	0.36	0.52	0.15	0.04	0.09	0.011
NPS + M	15.4	2.31	0.53	0.61	0.15	0.04	0.13	0.008

Note: Yields based on a 4 square yard sample; chemical analyses averages of duplicates and expressed on oven dry basis.

The nutrient content of grain is influenced to a large extent by the preceding legume crop. Crops following the fertilized legumes contain considerably larger amounts of protein than do crops grown on fallow. Some of the mineral constituents increased while others decreased but to a lesser degree than the protein content.

2. Rat Feeding Trials - Three feeding trials were conducted in 1956 using rats to evaluate the nutritive value of Breton wheat.

In feeding trial number 1 reported in Table 4, rats were fed a diet of wheat that was on rotation with legumes. There were three main reasons for using the grain from the six plots.

(a) The nutritive value of the fertilized grain was expected to show the greatest difference since Renner (52) found that the protein content and amino acid proportions were increased compared to the check.

(b) One feeding trial would indicate the growth difference, if any, to be expected in subsequent work.

(c) The data obtained would be used for comparison with wheat grown after fallow even if there were no differences in rat gains due to fertilization.

Some of the headings used in Table 4 and subsequent tables dealing with feeding trials are not in common use. Therefore, the following formulae show the derivation of some of the factors involved.

(a) $$\text{\% Dig. Energy} = \frac{\text{Gross energy} - \text{Fecal energy}}{\text{Total energy of feed}} \times 100$$

(b) $$\text{\% Dig. N} = \frac{\text{Feed N} - \text{Fecal N}}{\text{Feed N}} \times 100$$

(c) $$\text{N Retained} = \frac{\text{Feed N} - (\text{Fecal N} + \text{Urinary N})}{\text{Feed N}} \times 100$$

Table No. 4

FEEDING TRIAL NO. I, BRETON WHEAT AFTER LEGUMES - 1956

Treatment	Yield bu/ac.	Average rat gain /7 days gms.	Av. food consumed /7 days gms.	Av. food /gm gain gms.	Digestible Energy %	Digestible Nitrogen %	Nitrogen retained %	Digestible Energy/gm dig. N	lbs. of N retained/ acre lbs.
Wheat after legumes									
Check	13.8	13.8	57.0	4.1	88.0	87.5	44.2	181	7.8
L	13.4	12.9	54.1	4.2	83.5	89.3	38.3	157	7.6
Mean of low yielding	13.6	13.4		4.2	85.8	88.4			7.7
M	34.8	17.8	69.4	3.9	87.8	87.4	47.2	163	23.8
NS	38.7	21.5*	60.5	2.8*	85.8	84.7	42.4	163	23.3
NPKS	33.6	17.1	62.6	3.7	89.3	87.9	46.9	169	22.1
NPS + M	42.3	20.4*	68.1	3.3	86.6	86.2	45.1	160	26.6
Mean of high yielding	37.3	19.2		3.4	87.4	86.6			24.0
* L.S.D. P=0.05		5.1		0.9					

Rat data average of 4 replicates.

(d) Dig. Energy/gm. Dig. N = $\frac{\text{Gross energy - fecal energy}}{\text{Feed N - Fecal N}}$

(e) lbs. of N retained/ac. = yield bu./ac. x 60 lbs./bu. x $\frac{\text{\%N(grain)}}{100} \times \frac{\text{\%N retained}}{100}$

In table 4 the following conclusions appear to be justified.

(a) Fertilizer application to wheat plots resulted in an increase in rat gain from a low yielding mean of 13.4 gms. to a high yielding mean of 19.2 gms.

(b) Ammonium sulfate fertilization increased both rat gains and food efficiency significantly at the 5 per cent level, while NPS + M increased only rat gains significantly.

(c) Fertilizer applications to grain resulted in slight increases in per cent digestible energy and slight decreases in per cent digestible nitrogen. The small changes are mainly due to variability in food consumption between treatments.

(d) The application of all fertilizers decreased the digestible energy per gram of digestible nitrogen ratio. The fertiliser treatments have resulted in a greater proportion of nitrogen than energy being retained compared to the control treatment.

(e) The yield of plots influences the calculated value of pounds of nitrogen retained per acre with the NPS + M treatment showing the greatest increase.

Feeding trial number II presented in Table 5 was intended to compare wheat after legumes with wheat after fallow. The following conclusions appear justified.

Table No. 5

FEEDING TRIAL NO. II COMPARING BRETON WHEATS 1956

Treatment	Yield bu/ac.	Average rat gain /7 days gms.	Av. food consumed /7 days gms.	Av. food /gm gain gms.	Digestible Energy %	Digestible Nitrogen %	Nitrogen retained %	Digestible Energy/gm dig. N	lbs. of N retained/ acre lbs.
Wheat after legumes									
Check	13.8	12.0	53.3	4.4	87.4	86.4	36.1	182	6.3
NS	38.7	16.1	62.3	3.9	87.2	86.8	43.2	160	27.8
NPKS	33.6	19.4*	68.5	3.5	85.5	82.7	41.4	172	19.4
NPS + M	42.3	11.6	53.8	4.6	87.7	86.0	39.6	162	23.3
Mean of high yielding	38.2	15.7		4.0	86.8	85.2			23.5
Wheat after fallow									
Check	8.6	10.7	52.1	4.9	85.5	81.9	35.4	204	3.6
NS	7.1	8.6	50.3	5.8	85.3	83.2	36.1	197	3.1
NPKS	10.3	7.6	47.6	6.3	86.8	81.6	34.3	218	4.0
NPS + M	14.5	14.0	57.2	4.1	87.2	86.8	40.5	181	7.5
Mean of high yielding	10.6	10.1		5.4	86.4	83.9			4.9

*L.S.D. P = 0.05 7.0
(wheat after fallow)

Rat data average of 4 replicates.

(a) Wheat after legumes gave higher rat gains and better feed efficiency than wheat after fallow.

(b) The application of fertilizers to wheat after legumes resulted in a greater increase in rat gain than that of wheat after fallow when comparing the high yielding means of the two wheat crops.

(c) NPS + M of wheat after fallow gave the largest increase in rat gain of the fertilizer treatments on that crop.

(d) The poor yield of wheat after fallow as well as very low fertilizer response compared to the 26 year average at Breton (2), has resulted in low calculated values of pounds of nitrogen retained per acre. This low value is also influenced by the low nitrogen content of wheat after fallow compared to wheat after legumes in Table 2.

Feeding trial number III reported in Table 6, was intended to further compare wheat after legumes with wheat after fallow. In this trial, the manure treatment of both wheat crops was included and, because of lack of facilities, NPKS of both wheat crops was omitted. The NPS + M treatment of wheat after legumes was repeated because of lack of agreement in rat gains between feeding trials I and II.

(a) The NPS + M treatment on wheat after legumes showed the largest rat gain in this feeding trial, but, due to variability between replicates, it did not prove to be a significant difference.

(b) The preceding legume crop again improved the feeding value of wheat more than the application of fertilizers as is shown in rat gains and food/gm. gain.

Table No. 6 FEEDING TRIAL NO. III - A FURTHER COMPARISON OF BRETON WHEATS 1956

Treatment	Yield bu/ac.	Average rat gain /7 days gms.	Av. food consumed /7 days gms.	Av. food /gm gain gms.	Digestible Energy %	Digestible Nitrogen %	Nitrogen retained %	Digestible Energy/gm dig. N	Lbs of N retained/ acre lbs.
Wheat after legumes									
Check	13.8	11.4	55.8	4.9	86.9	86.2	33.0	181	5.8
M	34.8	14.7	64.6	4.4	87.0	85.5	37.6	164	18.9
NS	38.7	16.7	66.2	4.0	86.3	85.2	42.7*	161	23.5
NPS + M	42.3	19.3	72.7	3.8	85.5	84.0	41.8*	162	24.6
Mean of high yielding	38.6	16.9		4.0	86.3	84.9	40.7		22.3
Wheat after fallow									
Check	8.6	8.0	54.2	6.8	85.3	82.2	34.7	202	3.5
L	10.8	11.2	59.8	5.3	85.3	80.8	41.1*	203	5.6
Mean of low yielding	9.7	9.6		6.1	85.3	81.5	37.9		4.5
M	8.3	15.6	65.4	4.2	87.4	87.4	47.1*	186	4.3
NS	7.1	7.7	51.4	6.7	85.8	82.4	38.0	200	3.2
Mean of high yielding	7.7	11.6		5.4	86.6	84.9	42.6		3.8
* L.S.D. P = 0.05							7.5		

Rat data average of 4 replicates.

(c) Manure application to wheat after fallow increased rat gain from 8.0 gms. on the check treatment to 15.6 gms. This increase in rat gain is equal to any obtained with fertilized wheat after legumes in feeding trial II.

(d) The per cent nitrogen retained in the rat body has been significantly increased by the NS and NPS + M treatments of wheat after legumes, and lime and manure treatments of wheat after fallow.

(e) The ratio, digestible energy/gm. of digestible nitrogen, has been decreased by all fertilized treatments of wheat after legumes, but in wheat after fallow only the manure treatment has resulted in a decrease in this ratio. This again shows the beneficial effect of manure application to wheat after fallow.

B. <u>1957 Breton Grains</u>

I <u>Yield and Chemical Composition</u> - Fertilizers are applied every two years on the Breton plots. Since these applications were made in 1956, the 1957 fertilizer results are a residual effect.

Yield results of wheat after legumes in 1957 (Table 7) are similar to those of 1956 (Table 2) while the wheat yield after fallow in 1957 is higher than the 1956 results. The yield of the check plot and the increase in yield from manure and inorganic fertilizers in the 1957 wheat after fallow plots are considerably greater than the 26 year average results reported from the same plots (2). The largest increase in yield of 1957 wheat after fallow is from the application of

Table No. 7 YIELD AND CHEMICAL COMPOSITION OF 1957 BRETON WHEAT

Treatment	Yield bu/ac.	%N	%P	%K	%S	%Ca	%Mg	%Na
Wheat after legumes								
Check	11.6	2.82	0.49	0.37	0.10	0.04	0.16	0.010
L	12.3	2.84	0.51	0.34	0.07	0.04	0.12	0.012
M	21.2	2.68	0.52	0.30	0.07	0.04	0.14	0.008
NS	36.5	2.72	0.34	0.29	0.13	0.03	0.12	0.010
NPKS	38.5	2.42	0.36	0.32	0.12	0.03	0.12	0.014
NPS + M	30.0	2.82	0.39	0.34	0.12	0.03	0.13	0.009
Wheat after fallow								
Check	17.0	2.03	0.41	0.38	0.13	0.04	0.14	0.026
L	17.8	2.00	0.40	0.36	0.09	0.04	0.18	0.014
M	43.8	2.26	0.36	0.34	0.09	0.04	0.13	0.014
NS	25.8	2.12	0.36	0.34	0.09	0.03	0.11	0.008
NPKS	33.9	2.03	0.34	0.36	0.08	0.04	0.12	0.013
NPS + M	40.7	2.35	0.39	0.33	0.10	0.02	0.13	0.008

Note: Yields based on a 4 square yard sample; chemical analyses averages of duplicates and expressed on oven dry basis.

manure. The manure application was made in the spring of 1957 which may have improved the physical properties of the soil thus giving a better seedbed and increased yields. From the table it is evident that yields of wheat after fallow in general are higher than that of wheat after legumes, but there is a marked increase in yield above the long time average (2) on both crops due to the application of fertilizers.

In general, the mineral and protein content of 1957 Breton wheat reported in Table 7 was decreased by the application of nitrogen and sulfur fertilizers. The nitrogen content of wheat after legumes shows a decrease due to the application of M, NS and NPKS, but was unaffected by the application of L or NPS + M. The nitrogen content of wheat after legumes is higher on all plots than on the corresponding 1956 plots. On wheat after fallow the nitrogen content was similar in both years. The seasonal variation in protein content agrees with the findings of Ellis (11) in Manitoba. On wheat after fallow, only the application of manure resulted in an increase in nitrogen percentage. The phosphorus percentage of both wheat crops is similar to those reported by Morrison (39). The application of NS, NPKS and NPS + M to wheat after legumes and wheat after fallow caused a slight decrease in phosphorus content. This decrease in phosphorus content resulting from the application of nitrogen fertilizers agrees with results reported by Rennie (53). In addition, he found greater changes in phosphorus content between seasons than from the application of fertilizers, which is also evident when comparing 1957 data with 1956 data (Table 2). Similarly the application of manure to wheat after fallow (Table 7) decreased the phosphorus content. The lime treatment had very little effect on the phosphorus or nitrogen content of either wheat after legumes or wheat after fallow.

Of the mineral constituents in Table 7, only the sulfur content of wheat after legumes shows an increase due to fertilization. The difference in mineral composition between seasons is equal to, and in some cases greater than the difference due to the preceding legume crop or fertilization.

The yield data and results of chemical analyses of 1957 Breton barley, the first and third year after legumes are reported in Table 8. The yield of both barley crops shows a large increase due to the application of fertilizers with the exception of the lime treatment. The yields and increases are similar to the 26 year average, reported on the Breton barley (2).

The mineral content of fertilized plots from both barley crops was markedly decreased compared to the unfertilized and lime plots. The nitrogen, phosphorus and potassium content of first year barley after legumes are decreased by the same fertilization, namely; NS, NPKS and NPS + M. The same trend is evident in third year barley after legumes, with the exception that the manure treatment decreased the nitrogen and phosphorus content but not the per cent potassium. Considering all treatments, the first year barley is higher in per cent nitrogen than the third year barley which may be due to the third year barley occupying the least favorable position of the three grain crops in the rotation (43). Of the nutrients determined in the 1957 barley crops, only the sulfur content increased by the application of nitrogen and sulfur containing fertilizers. Morrison (39) reports 0.15 per cent as the sulfur percentage of barley which is slightly higher than the values reported in Table 8. The increase in per cent sulfur reported here is similar to results reported by Peters (46) and Hoff (20).

Table No. 8 YIELD AND CHEMICAL COMPOSITION OF 1957 BRETON BARLEY

Treatment	Yield bu/ac.	%N	%P	%K	%S	%Ca	%Mg	%Na
First year barley after legumes								
Check	9.4	2.36	0.54	0.60	0.07	0.03	0.14	0.020
L	12.6	2.38	0.62	0.62	0.05	0.04	0.14	0.016
M	41.3	2.36	0.54	0.55	0.08	0.03	0.14	0.014
NS	54.0	2.16	0.26	0.44	0.12	0.04	0.12	0.016
NPKS	69.7	1.96	0.34	0.46	0.11	0;04	0.13	0.018
NPS + M	65.7	2.44	0.44	0.47	0.11	0.04	0.14	0.015
Third year barley after legumes								
Check	14.4	2.02	0.56	0.60	0.08	0.04	0.16	0.016
L	14.3	1.82	0.62	0.59	0.08	0.04	0.14	0.016
M	50.9	1.62	0.36	0.50	0.08	0.04	0.14	0.014
NS	45.3	1.57	0.36	0.42	0.11	0.03	0.12	0.014
NPKS	40.6	1.48	0.39	0.48	0.10	0.04	0.12	0.014
NPS + M	65.5	1.84	0.53	0.47	0.12	0.04	0.13	0.013

Note: Yields based on a 4 square yard sample; chemical analyses averages of duplicates and expressed on oven dry basis.

2. Rat Feeding Trials - One feeding trial was conducted using 1957 wheat after legumes and wheat after fallow. The results are reported in Table 9. The number of replicates was increased to overcome some of the variation between replicates that existed in the 1956 feeding trials. Due to limited cage facilities, only one or two replicates of the rats were on test during any one test period. The feeding trial required five weeks to complete the six replications of each treatment.

From Table 9 the following conclusions appear justified.

(a) When comparing low and high yielding means of rat gains on wheat after legumes and wheat after fallow, there is no difference due to the application of fertilizers.

(b) The application of NPS + M to both wheat crops resulted in the largest increase in rat gains.

(c) Rat gains were increased when fed wheat after legumes compared to wheat after fallow.

(d) The average food consumption by the rats was greater with wheat after legumes than wheat after fallow.

(e) The average food consumed per gram of rat gain shows that wheat after legumes is superior to wheat after fallow, but again there is little or no difference due to fertilizer application.

(f) Digestible energy and digestible nitrogen appeared to decrease in fertilized wheat after legumes (low and high yielding means) while these digestibilities increased in wheat after fallow.

Table No. 9

FEEDING TRIAL NO. IV - COMPARING BRETON WHEATS - 1957

Treatment	Yield bu/ac.	Average rat gain / 7 days gms.	Av. food consumed /7 days gms.	Av. food /gm gain gms.	Digestible Energy %	Digestible Nitrogen %	Nitrogen retained %	Digestible Energy/gm dig. N	Lbs. of N retained/ acre lbs.
Wheat after legumes									
Check	11.6	21.9	68.3	3.2	87.6	88.4	46.4	133	9.1
L	12.3	18.4	63.5	3.6	87.6	89.5	46.0	131	9.6
Mean of low yielding	12.0	20.2		3.4	87.6	89.0	46.2		9.4
M	21.2	21.4	70.4	3.3	87.6	89.1	49.4	140	16.8
S	36.5	18.6	62.8	3.6	85.4	84.2	46.5	142	27.7
NPKS	38.5	18.2	60.4	3.4	86.5	84.1	47.6	161	26.6
NPS + M	30.0	23.7	68.8	3.0	87.0	86.9	51.3	135	26.0
Mean of high yielding	31.5	20.5		3.3	86.6	86.1	48.7		24.3
Wheat after fallow									
Check	17.0	13.6	55.5	4.1	84.7	78.1	40.6	199	8.4
L	17.8	16.1	56.1	3.5	85.4	80.5	45.2*	199	9.7
Mean of low yielding	17.4	14.8		3.8	85.0	79.3	42.9		9.0
M	43.8	15.3	58.0	4.3	87.7	86.1	52.8*	168	31.4
NS	25.8	15.2	58.1	3.9	86.0	81.7	45.2*	186	14.8
NPKS	33.9	11.1	50.0	4.6	87.0	81.8	45.4*	195	18.7
NPS + M	40.7	17.5	59.0	3.4	87.6	85.2	48.7*	167	27.9
Mean of high yielding	36.0	14.8		4.1	87.1	83.7	48.0		23.2

* L.S.D. P = 0.05

Rat data average of 6 replicates.

(g) When comparing the mean values of both wheat crops, the per cent nitrogen retained in the rat body increased when fed grain from fertilized plots. The increase in per cent nitrogen retained by the rats fed wheat after fallow was significant at the 5 per cent level. This is partly explainable on the basis of differences in feed consumption coupled with differences in per cent nitrogen in this grain as well as differences in nitrogen excreted.

(h) There was a slight decrease in nitrogen retention from the mean of the high yielding plots of wheat after legumes to the corresponding value of wheat after fallow. The application of fertilizers (not lime) resulted in a large increase in this calculated value.

SUMMARY AND CONCLUSIONS

A. Yield and Chemical Composition of Breton Grains - The yield of wheat and barley was increased both by the application of fertilizers and by growing legumes in rotation with these grains.

1. The preceding legume crop changed the nitrogen percentage more than the application of fertilizers during the two years under investigation. Seasonal variation has caused marked changes in the nitrogen content, especially on wheat after legumes. The wheat after legumes in 1957 was higher in per cent nitrogen than the corresponding crop in 1956. The per cent nitrogen reported is in general the same as that reported by Morrison (39) for the average of all types of wheat.

2. The application of NS, NPKS and NPS + M has resulted in more consistent decreases in the phosphorus content of both wheat and barley during 1956 than either M alone or lime. The effect of seasonal variations on the phosphorus content of Breton grains was small compared to that due to fertilization. The values reported for phosphorus are slightly lower than Morrison (39) states for grains of the northern plains.

3. Fertilizer application generally decreased the per cent sodium and potassium of wheat and barley in the two years, 1956 and 1957 while the calcium content remained relatively unchanged. The magnesium content decreased with the application of NS, NPKS and NPS + M.

4. Application of sulfur containing fertilizers (NS, NPKS and NPS + M) generally resulted in an increase in the sulfur content of wheat and barley after legumes. The application of fertilizers to wheat after fallow had no effect on the per cent sulfur in the 1956 crop but decreased the per cent sulfur in the 1957 crop. This decrease in per cent

sulfur content in the 1957 wheat after fallow crop is unexplainable on the basis of a calculation of the pounds of sulfur removed by the crop.

5. The application of lime to the Grey Wooded soil at Breton has not affected the yield and chemical composition of wheat and barley crops.

The 1956 yield of wheat after fallow reported in Table 2 is considerably lower than either the 26 year average of the same plots (2) or data reported by Newton et al. (43). It is possible that an error was made in calculating these yields. However, there is no possible way of checking the accuracy of these yields so they remain questionable. If, in fact, these are the correct yields, then 1956 was an unusual year and the results of the feeding trial with wheat after fallow may not be truly representative of the grain from these plots.

B. Rat Feeding Trials with Breton Wheat - Plate I is an isometric graphical presentation of average rat gains for the three feeding trials conducted in 1956.

1. The manure application to wheat after fallow resulted in the greatest increase in rat gains.

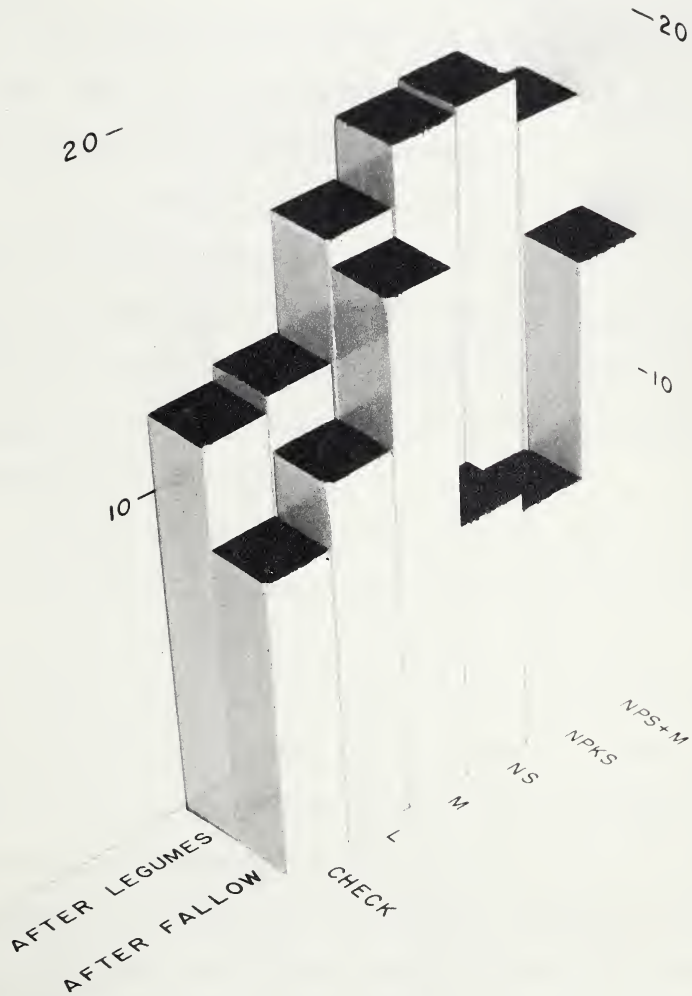
2. When legumes are included in the rotation, the application of sulfur containing fertilizers appears to increase the nutritive value of the wheat.

3. The nutritive value of both wheat crops, as measured by rat gains showed no difference due to the application of lime. This is consistent with yield and chemical composition.

Plate 1

AVERAGE RAT GAINS - 1956 BRETON WHEAT

Grams per Week



The results of the 1957 feeding trial are presented graphically in Plate II.

1. There is very little effect of fertilization on rat gains.

2. The application of manure both alone and with NPS + M on wheat after fallow shows a slight increase in rat gain similar to that in 1956 data.

3. The nutritive value of wheat is improved when preceded by a legume crop. The application of fertilizers resulted in only a slight additional improvement in nutritional value.

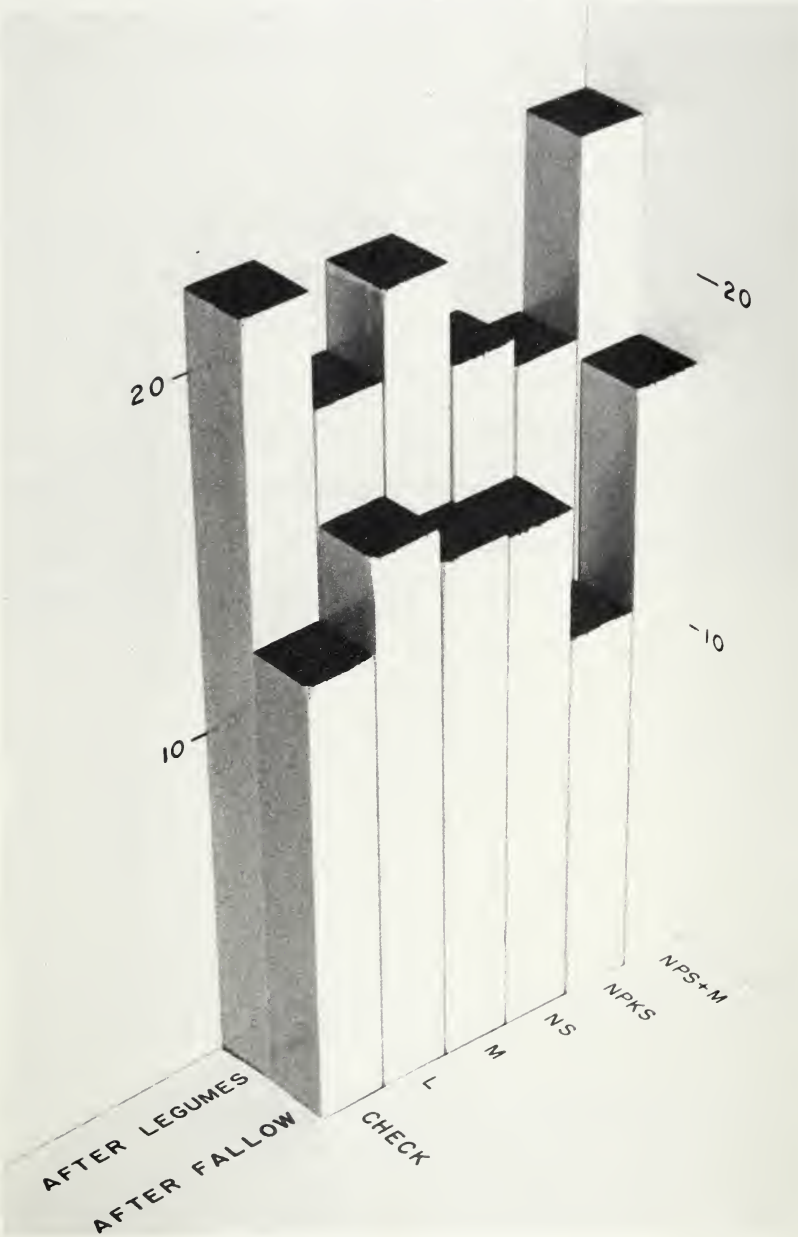
The rat feeding trials conducted in this investigation were preliminary studies and interpolation to other classes of animals requires additional investigation. Although the rat is a monogastric animal similar to swine (31), there still remains a great deal of variability between rats and any increase in gains should be interpreted as trends to be followed in future studies.

Factors affecting rat growth are not confined to the protein content of the grain as was studied and herein reported. One undesirable factor evident in the 1957 feeding trial was measuring rat growth at different times. Since the rats were under very strictly controlled conditions, then a slight change of temperature for example may cause variations in rat growth. Another factor which may affect rat growth either directly or indirectly is the absence of trace elements. As a specific example molybdenum is a trace element important in nitrogen reduction within the plant and as a result it can affect the protein of crops. Although this was not studied with regard to the Breton plots, this could conceivably be another factor affecting rat growth.

Plate 11

AVERAGE RAT GAINS - 1957 BRETON WHEAT

Grams per Week



Investigations initiated to study factors that may be responsible for changes in the nutritive value of grains would necessitate a systematic long term approach. Since grain from the Breton plots is available in large quantities, then this is one place to start such a long term investigation. The feeding trials could be used to study such factors as amino acids, minerals and vitamins by a process of elimination. For example, if minerals were found to be the most deficient nutrient in grain as measured by decreased rat growth, then, by means of a series of feeding trials, the most limiting minerals could be determined. The same could apply to amino acids or vitamins. The treatments from the Breton plots would have to be carefully selected. The project could be conducted by the department with graduate or student assistance wherever possible. The eventual outcome would likely be a clearer understanding of factors affecting the nutritive value of Breton grains.

REFERENCES

1. Alberta Advisory Fertilizer Committee. Annual compilation of data. 1956.

2. Alberta Advisory Fertilizer Committee. Annual compilation of data. 1957.

3. ALBRITTON, D. J., and R. ELLIS, Jr. Influence of organic matter and certain anions on the yield and phosphorus content of oats. Agron. J. 49: 410-412. 1957.

4. Anonymous. Oxygen bomb calorimetry and oxygen bomb combustion methods. Parr Manual No. 120, Parr Instrument Co., Moline, Ill. 1948.

5. BAINS, G. S. Effect of commercial fertilizers and green manures on yield and nutritive value of wheat. I. Nutritive value with respect to total phosphorus, phytic phosphorus, non-phytic phosphorus, and calcium content of grain. Cereal Chem. 26: 317-325. 1949.

6. BAINS, G. S. Effect of commercial fertilizers and green manures on yield and nutritive value of wheat. II. Nutritive value with respect to general composition, thiamine, nicotinic acid, and biological value of the protein of grain. Cereal Chem. 30: 139-145. 1953.

7. BELL, J. M. A comparison of peroxide and oxygen bomb calorimetry of feedstuff. Can. J. Agr. Sci. 35: 366-370. 1955.

8. BROWN, R. A., and M. STURTEVANT. The vitamin requirements of growing rats. Vitamins and Hormones. 7: 171-197. 1949.

9. CRAMPTON, E. W., and L. C. RAYMOND. Fertilization of permanent pastures for steer grazing. McGill Univ. Tech. Bull. 13. 1934.

10. DEAN, L. A. Fixation of soil phosphorus. Advances in Agronomy. I: 391-409. 1949.

11. ELLIS, J. H. Report on effect of certain cultural and fertility practices on the protein content of wheat. Sci. Agr. 23: 474-495. 1943.

12. ELLIS, B. G., C. J. KNAUSS, and F. W. SMITH. Nutrient content of corn as related to fertilizer application and soil fertility. Agron. J. 48: 455-459. 1956.

13. GAREAU, L. Some effects of commercial fertilizers on yield and quality of forage crops in Alberta. M.Sc. Thesis, Univ. of Alberta. 1955.

14. GEIGER, E., I. E. RAWI, and H. V. THOMAS. Experiments with intermittent feeding of protein to rats. J. Nutrition. 56: 273-283. 1955.

15. GIESEKING, J. E., N. I. SNIDER, and C. N. GETZ. Destruction of organic matter in plant material by use of nitric and perchloric acids. Ind. Eng. Chem. Anal. Ed. 7: 185-186. 1935.

16. HARPER, A. E., D. A. BENTON, and C. A. ELVEHJEM. L-leucine, an isoleucine antagonist in the rat. Arch. Biochem. and Biophys. 57: 1-8 1955.

17. HARPER, A. E., M. E. WINJE, D. A. BENTON, and C. A. ELVEHJEM. Effect of amino acid supplements on growth and fat deposition in the liver of rata fed polished rice. J. Nutrition. 56: 187-198. 1955.

18. HEDLIN, R. A., R. E. SMITH, and F. P. LECLAIRE. Effect of crop residues and fertilizer treatments on yield and protein content of wheat. Can. J. Soil Sci. 37: 34-40. 1957.

19. HILL, K. W. Wheat yields and soil fertility on Canadian prairies after a half century of farming. Soil Sci. Soc. Amer. Proc. 18: 182-184. 1954.

20. HOFF, D. J. Fertility studies with radioactive and non-radioactive sulfur on grey wooded soils of Alberta. M.Sc. Thesis, Univ. of Alberta. 1953.

21. HUPPERT, V. Nitrogen fertilization and the protein content of cereals. Limburgerhof Western Germany. Mitt. d. Deutsch. Landio. Ges, 1957, Vol. 72. No. 15: 359. (Abstracted Agr. Rev. No. 1. 3: 54. 1957.)

22. HUTCHINSON, J. B., T. MORAN, and J. PACE. Nutritive value of the protein of white and wholewheat bread in relation to the growth of rats. Proc. Roy. Soc. B. (London). 145: 270-279. 1956.

23. JELINEK, B., M. C. KATAYAMA, and A. E. HARPER. The inadequacy of unmodified potato starch as dietary carbohydrates for the albino rat. Can. J. Med. Sci. 30: 447-452. 1952.

24. KASTINGS. R. Sulfur-containing amino acids of wheat protein. M.Sc. Thesis, Univ. of Alberta. 1948.

25. KOEHLER, F. E., and W. A. ALBRECHT. Biosynthesis of amino acids according to soil fertility. Missouri Agr. Expt. Sta. J. No. 1342. 1953.

26. LAWTON, K., and J. E. DAVIS. The effect of liming on the utilization of soil and fertilizer phosphorus by several crops grown on acid organic soils. Soil Sci. Soc. Amer. Proc. 20: 522-526. 1956.

27. LIGHT, R. F., and C. N. FREY. The nutritive value of wheat and wholewheat bread. Cereal Chem. 20: 645-660. 1943.

28. LONG, O. H., and C. D. SHERBAKOFF. Effect of nitrogen on yield and quality of wheat. Agron. J. 43: 320-321. 1951.

29. LYND, J. Q., F. GRAYBILL, and R. TOTUSEK. Grazing trial evaluations using paired pastures with yearling steers. Agron. J. 49: 488-492. 1957.

30. MacLEAN, A. J., R. F. BISHOP, and L. E. LUTWICK. Fertility studies on soil types. 111. Phosphorus supply and requirement as shown by greenhouse studies and laboratory tests. Can. J. Agr. Sci. 33: 330-343. 1953.

31. MAYNARD, L. A., and J. K. LOOSLI. Animal nutrition. 4th Ed. McGraw-Hill Book Co., Toronto, Ont. 1956.

32. McELROY, L. W., and H. H. DRAPER. Feeding grains of different protein content to growing pigs. Sci. Agr. 29: 579-583. 1949.

33. McELROY, L. W., W. W. LOBAY, and R. D. SINCLAIR. Feeding grains of different protein content. J. Animal Sci. 7: 494-500. 1948.

34. McLEAN, E. O. Plant growth and uptake of nutrients as influenced by levels of nitrogen. Soil Sci. Soc. Amer. Proc. 21: 219-222. 1957.

35. MILLER, C. E. Soil fertility, pp. 396-422. John Wiley and Sons Inc., New York. 1955.

36. MITCHELL, H. H., and D. B. SMUTS. The amino acid deficiencies of beef, wheat, corn, oats, and soybeans for growth in the white rat. J. Biol. Chem. 95: 263. 1932.

37. MITCHELL, J. A review of tracer studies in Saskatchewan on the utilization of phosphates by grain crops. J. Soil Sci. 8: 73-85. 1957.

38. MOORE, D. G., O. J. ATLOE, and C. I. RICH. Phosphate fertilized recovered slowly in certain soils. Crops and Soils. Dec. p. 24. 1956.

39. MORRISON, F. B. Feeds and feeding. 22nd Ed. The Morrision Pub. Co., Ithaca, New York. 1956.

40. MURPHY, H. F. Effect of fertilizers on the yield and composition of wheat. J. Amer. Soc. Agron. 22: 765-770. 1930.

41. MURRAY, H. C. The biological value of the protein of field pea products with a comparison of several methods used for this determination. J. Nutrition. 35: 257-267. 1948.

42. MURRAY, H. C., S. KELLENBERGER, and G. A. ARMBUSTER. Protein efficiency and methionine content of Alaska peas grown with the use of various fertilizers. Agron. J. 44: 252-253. 1952.

43. NEWTON, J. D., A. S. WARD, and C. F. BENTLEY. Wooded soils and their management. Bull. No. 21, 4th Ed. (Revised). Univ. of Alberta. 1948.

44. Official Methods of Analysis of the Association of Official Agricultural Chemists. Washington 4, D.C. 8th Ed. 1955.

45. PAWLUK, S. The exchangeable cation status of some west central Alberta soils. M.Sc. Thesis, Univ. of Alberta. 1955.

46. PETERS, T. W. The effect of fertilizers on the sulfur content of soil and crop plants. M.Sc. Thesis, Univ. of Alberta. 1942.

47. PETERSON, C. F., C. E. LAMPMAN, D. W. BOLIN, and O. E. STAMBERG. Methionine deficiency of Alaska field peas for chick growth. Poultry Sci. 23: 287-293. 1944.

48. PETERSON, H. B. Effect of nitrogen fertilizer on yields and protein content of winter wheat in Utah. Utah Agr. Expt. Sta. Bull. 353. 1952.

49. POWERS, W. L. The role of sulfur in plant nutrition. J. Amer. Soc. Agron. 22: 371-373. 1930.

50. PRINCE, A. L. Determination of total ammonia and nitrate in soils. Soil Sci. 59: 47-52. 1945.

51. RENDIG, V. V. Sulfur and nitrogen composition of fertilized and unfertilized alfalfa grown on sulfur-deficient soil. Proc. Soil Sci. Soc. Amer. 20: 237-240. 1956.

52. RENNER, R., C. F. BENTLEY, and L. W. McELROY. Nine essential amino acids in the protein of wheat and barley grown on sulfur-deficient soil. Soil Sci. Soc. Amer. Proc. 17: 270-273. 1953.

53. RENNIE, D. A. Variations in percentage phosphorus and protein content of wheat, as induced by fertilizer treatment, soil type and season. Can. J. Agr. Sci. 36: 491-504. 1956.

54. ROSENBERG, H. R., and E. L. ROHDENBERG. The fortification of bread with lysine. II. The nutritional value of fortified bread. Arch. Biochem. and Biophys. 37: 461-468. 1952.

55. ROSENBERG, H. R., E. L. ROHDENBERG, and J. T. BALDINI. The fortification of bread with lysine. III. Supplementation with essential amino acids. Arch. Biochem. and Biophys. 49: 263-267. 1954.

56. RUSSELL, J. S., C. W. BOURG, and H. F. RHOADES. Effect of nitrogen fertilizer on the nitrogen, phosphorus, and cation contents of bromegrass. Soil Sci. Soc. Amer. Proc. 18: 292-296. 1954.

57. SIBBALD, I. R., R. T. BERG, and J. P. BOWLAND. Digestible energy in relation to food intake and nitrogen retention in the weanling rat. J. Nutrition. 59: 385-392. 1956.

58. SMITH, G. E., and W. A. ALBRECHT. Feed efficiency in terms of biological assays of soil treatments. Soil Sci. Soc. Amer. Proc. 7: 322-330. 1942.

59. SURE, B. Influence of lysine, valine and threonine additions on the efficiency of the protein of wholewheat. Arch. Biochem. and Biophys. 39: 463-464. 1952.

60. VANDECAVEYE, S. C. Effects of soil type and fertilizer treatments on the chemical composition of certain forage and small grain crops. Soil Sci. Soc. Amer. Proc. 5: 107-125. 1940.

61. WEDIN, W. F., A. W. BURGER, and H. L. AHLGREN. Effect of soil type, fertilization, and stage of growth on yield chemical composition and biological value of ladino clover and alfalfa. Agron. J. 48: 147-152. 1956.

62. WEHUNT, R. L., M. STELLY, and W. O. COLLINS. Effect of sodium and potassium on concentration and crimson clover grown on Norfolk sandy loam at two residual potassium levels. Soil Sci. 83: 175-183. 1957.

63. WEIR, W. C., and V. V. RENDIG. Studies on the nutritive value for lambs of alfalfa hay grown in a low sulfur soil. J. Animal Sci. (Abstract) 11: 780-781. 1952.

64. WOODS, E., W. M. BEESON, and D. W. BOLIN. Field peas as a source of protein for growth. J. Nutrition. 26: 327-335. 1943.

Table No. 10 YIELD AND CHEMICAL COMPOSITION OF 1956 COOPERATIVE FERTILIZER TRIALS ON GRAIN

Treatment: Nutrients supplied, approx. lb./ac. N	P_2O_5	K_2O	Yield bu./ac. Absolute yield	Inc. over check	%N	%P	%K	%S	%Ca	%Mg	%Na
STEVENSON, H. Delia - Wheat - Dark Brown											
0	0		25.1	-	2.58	0.31	0.46	0.16	0.04	0.13	0.006
40	0			1.2	2.83	0.29	0.45	0.18	0.04	0.13	0.008
0	40			12.6	2.45	0.30	0.42	0.15	0.04	0.13	0.007
40	40			17.8	2.58	0.28	0.42	0.16	0.04	0.13	0.008
40	40	12.5		11.4	2.63	0.10	0.39	0.16	0.04	0.07	0.006
10	40			14.6	2.33	0.20	0.41	0.17	0.04	0.10	0.017
20	40			13.9	2.44	0.29	0.42	0.18	0.03	0.13	0.007
40	10			11.4	2.56	0.21	0.37	0.18	0.04	0.11	0.007
40	20			14.6	2.52	0.27	0.38	0.17	0.04	0.12	0.006
NIDDRIE, F. Olds - Barley - Grey Wooded											
0	0		42.6	-	1.63	0.46	0.59	0.11	0.06	0.14	0.009
40	0			16.8	2.01	0.45	0.64	0.16	0.06	0.14	0.007
0	40			1.9	1.72	0.43	0.64	0.12	0.06	0.13	0.012
40	40			2.0	2.05	0.43	0.60	0.16	0.07	0.14	0.009
40	40	12.5		-2.4	2.02	0.40	0.61	0.14	0.08	0.13	0.012
10	40			-2.4	1.78	0.37	0.62	0.13	0.06	0.12	0.008
20	40			0.8	1.91	0.36	0.66	0.14	0.07	0.11	0.008
40	10			-6.9	2.14	0.41	0.66	0.18	0.07	0.14	0.008
40	20			-5.0	2.11	0.42	0.66	0.17	0.08	0.12	0.007

APPENDIX

Table No. 10 continued

YIELD AND CHEMICAL COMPOSITION OF 1956 COOPERATIVE FERTILIZER TRIALS ON GRAIN

Treatment: Nutrients supplied, approx. lb./ac. N	P_2O_5	K_2O	Yield bu./ac. Absolute yield	Inc. over. check	%N	%P	%K	%S	%Ca	%Mg	%Na
SANDERS, R. Duhamel - Barley - Black											
0	0	0	11.5	-	1.63	0.28	0.58	0.13	0.06	0.11	0.008
80	0			7.3	2.28	0.25	0.54	0.16	0.04	0.11	0.009
0	40			4.8	1.54	0.27	0.58	0.10	0.04	0.10	0.008
80	40			20.4	2.08	0.30	0.53	0.16	0.04	0.12	0.011
80	40	12.5		22.8	1.95	0.33	0.54	0.12	0.03	0.12	0.007
HAGA, I. Vulcan - Wheat - Dark Brown											
0	0		16.6		1.99	0.37	0.45	0.13			0.010
55	0			23.6	2.24	0.33	0.36	0.14	0.03	0.14	0.012
47	24			21.1	2.12	0.34	0.42	0.09	0.03	0.14	0.007
41	0			24.1	2.08	0.32	0.41	0.14	0.03	0.14	0.008
50	0			15.4	1.88	0.34	0.44	0.15	0.03	0.15	0.008
33	24			15.4	1.95	0.34	0.40	0.12	0.03	0.14	0.017
MERCHANT, R. M. Coronation - Wheat - Dark Brown											
0	0		26.8		2.17	0.26	0.40	0.12	0.03	0.10	0.015
55	0			6.2	2.40	0.19	0.35	0.11	0.03	0.09	0.010
47	24			18.1	2.42	0.15	0.36	0.11	0.03	0.08	0.011
41	0			7.1	2.47	0.22	0.36	0.14	0.04	0.12	0.010
50	0			5.2	2.43	0.23	0.38	0.14	0.04	0.12	0.009
33	24			15.8	2.36	0.27	0.36	0.15	0.05	0.13	0.008

Table No. 10 continued — YIELD AND CHEMICAL COMPOSITION OF 1956 COOPERATIVE FERTILIZER TRIALS ON GRAIN

Treatment: Nutrients supplied, approx. lb./ac. N	P_2O_5	K_2O	Yield bu./ac. Absolute yield	Inc. over. check	%N	%P	%K	%S	%Ca	%Mg	%Na
MOSER, N Killam - Oats - Thin Black											
0	0		74.5		1.46	0.35	0.53	0.13	0.06	0.10	0.018
55	0			29.6	1.68	0.29	0.45	0.12	0.06	0.11	0.012
47	24			24.8	1.61	0.30	0.46	0.10	0.06	0.11	0.008
41	0			21.4	1.60	0.35	0.52	0.14	0.07	0.13	0.008
50	0			29.1	1.62	0.33	0.51	0.15	0.06	0.14	0.008
33	24			21.0	1.59	0.33	0.52	0.11	0.06	0.12	0.012

MICROKJELDAHL METHOD

Nitric-Perchloric Acid Disgestion Procedure for Mineral Constituents

Digestion of samples was on a rotary digestion apparatus described in an Aminco and Keogel catalogue*. A 110 volt rheostat was connected to digestion rack. The rheostat was calibrated to 100.

1. Weigh ± 2.4 g. into drying pans. (For hay, legume, grains).
2. Dry in oven for ± 4 hours or overnight at 110°C.
3. Weigh on the analytical balance and transfer without filter paper into 100 ml. Kjeldahl flasks. Add 2 or 3 glass beads to each Kjeldahl.
4. Add 25 cc. of nitric acid (dil 3:1), rinsing the neck of the Kjeldahl flask and leave for ± 2 hours with occasional shaking.
5. Leave on overnight on digester, rheostat at 40. Fume cupboard window closed. Do not put on glass fume hood.
6. Cool down in the morning and add 10 cc. of a mixture of 5 parts perchloric acid, 6 parts water, 2 parts nitric acid by volume. Put on aluminum foil, especially around the neck and part of bulb.
7. Put on digester rheostat set at 40 and try to bring the rheostat setting up to 75 in about 1 - 2 hours, taking care that the flasks do not bump and the liquid is boiling gently.
8. When all the nitric acid is evaporated and the white fumes of the perchloric acid begin to form, put on glass fume hood, set rheostat at 90. In order to evaporate last traces, set rheostat at 100. Do not take to dryness unless liquid is clear.

* American Instrument Co., Inc.
Silver Spring, Maryland, U.S.A. Bulletin 2271.

9. Cool. Add 5 cc. of conc. HCl, evaporate without glass fume hood. Start heating with rheostat at 70, then to remove last traces of perchloric set rheostat at 100.

10. Repeat 9.

11. Add 20 cc. of 1N HCl, loosen up residue using a stirring rod with policeman and filter through qualitative (No. 1 Whatman) filter paper into 200 cc. volumetric flask and make up to volume with distilled water.

PHOTOMETRIC DETERMINATION OF SODIUM

A. Standards

1. Prepare seven standard solutions of NaCl containing 0, 5, 10, 15, 20, 25, and 30 p.p.m. solution. All standard solutions contain the following concentrations of other ions:

35 p.p.m. P.

45 p.p.m. K.

7 p.p.m. Ca.

15 p.p.m. Mg.

The standards are then made up to 500 ml. with 0.1N HCl. The standard solutions thus obtained are similar in concentration of the various ions to the sample solution from ashing by the nitric-perchloric acid method.

2. Set the Beckman Model D.U. flame spectrophotometer with Model 9220 flame unit burning acetylene at 100 per cent transmission with the 30 p.p.m. standard.

(a) Fuel at 3.0 lbs. per sq. in.

(b) Oxygen at 10 lbs. per sq. in.

(c) Wavelength at 589.2 μ

(d) Slit width at 0.01

(e) Check switch at 0.1

(f) Blue phototube

(g) Phototube load resistor at 2

(h) Photomultiplier sensitivity at 4

(i) Zero suppression at 1

Determine the per cent transmission of the remaining six standards.

B. Determination of Unknown

1. The solution from the digestion mixture is transferred to spectrophotometer beakers and read in per cent transmission. A standard on either side of unknown is used to balance the spectrophotometer and to interpolate the concentration of unknown.

2. Calculation of per cent sodium as follows:

$$\%\text{Na} = \frac{\text{p.p.m.} \times \dfrac{\text{Volume of Digestion Solution (200)}}{1000}}{\text{sample wt} \times 1000} \times 100$$

$$= \frac{\text{p.p.m.} \times \dfrac{200}{1000}}{\text{Sample wt.} \times 1000} \times 100$$

$$= \frac{\text{p.p.m.} \times 0.02}{\text{Sample wt.}}$$

PHOTOMETRIC DETERMINATION OF POTASSIUM

A. Determination of Standards

1. Prepare seven standard solutions containing 0 (blank), 10, 20, 30, 40, 50, and 60. p.p.m. from an accurate 1000 p.p.m. standard.

2. All solutions contain the following concentrations of ions:

18 p.p.m. of P.

3 p.p.m. of Ca.

7 p.p.m. of Mg.

0.5 p.p.m. of Na.

3. The solutions were made up to 500 ml. with 0.1N HCl.

4. Set the Beckman Model D.U. spectrophotometer at 100 per cent transmission with the 60 p.p.m. standard. The following adjustments were found satisfactory:

(a) Fuel at 3.0 lbs. per sq. in.

(b) Oxygen at 10 lbs. per sq. in.

(c) Selector switch at 0.1

(d) Slit Width at 0.4

(e) Wave length at 771 μ.

(f) Red phototube

(g) Phototube load resistor at 3

Determine the per cent transmission of the remaining six standards.

B. Determination of Unknown

1. The solution obtained by ashing with nitric-perchloric acid method was diluted 1:1 with 0.1N HCl.

2. Transfer unknown solution to spectrophotometer beakers and per

cent transmission determined followed by two standards: one standard is next above the unknown; the second standard is next below the unknown.

3. From the per cent transmission of sample, the potassium concentration is found by interpolation between the two standards on either side of sample.

4. Calculation of per cent potassium in sample was as follows:

$$\%K = \frac{\frac{\text{p.p.m. x 2 (dilution of 1:1) x Vol. of Digestion solution x 100}}{1000}}{\text{Wt. of sample x 1000}}$$

$$\%K = \frac{\text{p.p.m. x 2 x } \frac{2}{10} \text{ x 100}}{\text{Wt. of sample x 1000}}$$

$$= \frac{\text{p.p.m. x 2 x 0.2}}{\text{Wt. of sample}} = \frac{\text{p.p.m. x 0.04}}{\text{Wt. of sample}}$$

Procedure for Mixing Supplements for 1956-57

Rat Feeding Trials

Minerals:

	Contents	Amount
A.	$CaCO_3$	534.0 gm.
B.	$MgCO_3$	25.0 gm.
C.	$MgSO_4$	16.0 gm.
D.	NaCl	69.0 gm.
E.	KCl	112.0 gm.
F.	KH_2PO_4	212.0 gm.
G.	Na_2HPO_4	800.0 gm.
H.	$FeSO_4$	60.0 gm.
I.	KI	0.08 gm.
J.	$MnSO_4 4H_2O$	16.2 gm.
K.	$Al_2(SO_4)_3 K_2SO_4 24H_2O$	0.17 gm.
L.	$CaSO_4$	0.90 gm.
M.	$ZnCl_2$	0.52 gm.

Minerals are included at a level of 4 per cent in all rations.

Use anhydrous chemicals unless otherwise stated, such as J. ($MnSO_4.4H_2O$). If the material is not anhydrous then calculate so as to take the water content into account. Mix on wax paper if possible as some chemicals stick to ordinary brown paper.

1. Mix A, B, C, F, G, H, and J thoroughly making sure all will pass through a 20 mesh sieve.

2. Mix D with I and K well (20 mesh).

3. Mix E with L and M well.

4. Mix the KCl mixture and NaCl mixture. Then mix with the others mixed under item 1.

5. Mix the whole salt mixture thoroughly making sure it will pass through a 20 mesh sieve and store in large brown paper bag (labelled). Large weights can be weighed on a triple beam balance but I, K, L, and M should be weighed on analytical balance.

Vitamin Mix

Contents	Weight
Inositol	10 gm.
Thiamine HCl	50 mg.
Riboflavin	100 mg.
Pyridoxine HCl	50 mg.
Niacin	50 mg.
Ca Pantothenate	250 mg.
P.A.B.A. (Para Amino Benzoic Acid)	3 gm.
α tocopherol acetate (vit E)	250 mg.
Menadione	20 mg.
Folic acid	20 mg.
Biotin	2 mg.
B_{12}	0.3 mg.
Vit. A Palmitate 10,000 I.U./gm.	60gm.
Dry Vit. D_2 8,000,000 I.U./lb.	6.81 gm

Make to 100 gm. with corn starch and include in all rations at a level of 1%.by weight.

Choline chloride solution 50 gm. made to 100 ml. with water. Add 0.4 ml./100 gm. of ration. i.e 0.2% choline chloride in ration.

1. Mix vitamins A and dry vit. D and inositol together on wax paper fairly well. Then spread this mixture over the paper.
2. Sprinkle the following over the mixture of item 1.: Thiamine, riboflavin, pyridoxine, niacin, Ca pantothenate, P.A.B.A., menadione, folic acid, biotin, and B_{12}. Mix this well, making sure it will pass through 20 mesh sieve.
3. Take a small quantity of the mixture from item 2 and place in a weighing pan. Weigh on analytical balance this pan plus mixture. By means of a glass rod place drops of tocopheral acetate on the mixture as it is an oily substance.
4. Mix the vitamins of item 3 to a fairly dry state and mix the whole mixture.
5. Add starch and mix and sieve through 20 mesh sieve. Store in refrigerator.
6. Mix choline chloride after the minerals and other vitamins have been mixed with the feed.

Feed and Supplement Mixing

Lysine mixed at 0.90% in a wheat ration such as was used. Total feed allowance for six rats per treatment was 1200 grams.

Ration used per 100 gms. total:

Grain	94.15
Minerals	4
Vitamins	1
Lysine	.90
	100.05

For 1,200 grams of feed:

Grain	1,129.8 gm.
Minerals	48.0 gm.
Vitamins	12.0 gm.
Lysine	10.20 gm.
	1,200.00 gm.

1. Mix the grain and minerals together fairly well.
2. Mix the vitamin mixture and lysine together.
3. Mix the mixture under item 1 and 2 together and put through 20 mesh sieve.
4. Mix 4.8 ml. of choline chloride solution with the mixture under item 3.
5. Place grain in container and store in the refrigerator to prevent deterioration. Do not mix a long time in advance of feeding trial.